Response to Office Action

Confirmation No.: 7662

We have been informed by the "Office Action" prepared by the examiner Ruth A. Davis, Art Unit 1651, that the claims of the patent application made by Tanil Zuhtu Kocagoz (and assigned to SALUBRIS, Inc.), numbered 10/683,565 filed on 10/10/2003 have been rejected in a non-final action, by letter dated March 07, 2006 and received at March 10, 2006. After reviewing the document, we have concluded without prejudice that the claims are rejected by misinterpretation of the definitions described in US patent numbered 6,579,694 belonging to Heifets et al. and US patent numbered 6,197,574 belonging to Miyomoto et al. We believe that upon review of the explanations provided below, it will be clear that the claims in our patent application are original and can not be rejected by referring to the claims described by Heifets et al and Miyomoto et al. We kindly request the acceptance of our claims as original for the reasons described in detail below.

General comments

In conventional mycobacterial culture media, the growth of mycobacteria is detected by visualization of colonies formed by mycobacteria, which generally requires 3 to 8 weeks. For those in the medical community attempting to determine the presence of the mycobacteria which causes tuberculosis, this is a long and potentially health threatening delay. To shorten the duration of time required for growth detection, several rapid mycobacterial culture systems using Middlebrook liquid medium have been developed. These systems have disadvantages, however, such as using radioactive substances, requiring expensive equipment and/or having difficulties in application. TK Medium, which is the subject of our patent application, is a new and original rapid mycobacterial culture medium which has many advantages over other rapid culture media and is completely unique in its method of detection and differentiation of the cultured organisms. TK Medium indicates mycobacterial growth by changing its original color from red to yellow. It also has the ability to differentiate the growth of contaminating organisms by changing from red to green, triggered by the growth of these organisms. These features are unique to TK Medium and are not present in any other rapid or conventional culture medium including the medium that is described by Heifets et al. (The media described by Miyomoto et al. is for growing the agents of food poisoning and can not grow mycobacteria.)

The medium described by Heifets et al. in the patent application numbered US 6,579,694 is not a rapid culture medium. There is no statement in any part of this patent document including its description and claims that this is a rapid culture medium or that it is used for rapid detection of mycobacterial growth. The medium described by Heifets et al. is basically a modification of Middlebrook medium, which replaces dry powder of bovine serum albumin with fresh animal serum. It should be noted that Heifets et al. obtained their patent by only this basic change although their newly described medium included all the substances that were previously described as Middlebrook Medium. All microbiological media contain common chemicals like proteins, carbohydrates and salts and sometimes one single chemical and even the preparation procedure makes a medium original and distinct from the others, as is the case of the medium described by Heifets et al.

The mechanism that leads to color change in TK Medium depends on two principles. The first principle is that the trigger of the color change which is the basis of TK Medium's rapid detection is caused by detecting the change in pH caused by mycobacterial growth. The original pH of TK Medium, which is 7.6, is decreased to 5.8 by mycobacterial growth. The medium contains a pH indicator, phenol red, which gives the red color to the medium at pH 7.6, and then turns to yellow when the pH is lowered to 5.8. The medium described by Heifets et al. is first of all a fixed pH culture medium having a pH between 6.0 and 6.2, where the pH is controlled very strictly not to change by the high concentration of phosphate salts (Monopotassium phosphate 1.5g/lt and Disodium phosphate 1.5g/lt) which are present in Middlebrook medium. As with most of the microbiological media, Middlebrook medium and the medium described by Heifets et al. are designed to keep the pH constant as much as possible during bacterial growth. The other important component, other than phosphate salts, of these media that keeps the pH constant is the albumin which is added as bovine serum albumin in Middlebrook medium and as the albumin that is contained in the animal serum in the medium described by Heifets et al. TK Medium does not contain any phosphate salt or bovine serum albumin. Contrary to Middlebrook Medium or the medium described by Heifets et al. TK Medium is designed to change its pH when there is mycobacterial growth. **There are no pH indicators in any mycobacterial culture medium described previously in any patent or practice.** The pH change in TK Medium is followed by the spectroscopic (color) change of TK Medium which indicates mycobacterial growth. Any pH indicator which works between pH 5.8 and 7.6 may be used in TK Medium. The preferred indicator is phenol red. Phenol red or any other pH indicator is not present in either Middlebrook medium or the medium described by Heifets et al. In the Office Communication, prepared by the examiner

Ruth A. Davis, Art Unit 1651, at page 6 the last sentence of the third paragraph states that "The medium further comprises ..., phenol red". We believe without prejudice that this is a misinterpretation of a sentence that contains "phenol red" included in the description of the patent US 6,579,694 belonging to Heifets et al. Phenol red is mentioned in this document in the section "Preparation of Samples for Drug Susceptibility Testing; NaOH-NALC Digestion-Decontamination Procedure with Neutralization" under the "Solutions required" second paragraph where the preparation of phosphate buffer is described. NaOH-NALC Digestion-Decontamination Procedure, described by Kubica et al. (Kubica GP, Dye WE, Cohn ML, Middlebrook G. Sputum digestion and decontamination with N-acetyl-L-cysteine-sodium hydroxide for culture of mycobacteria. 1963. Am. Rev. Respir. Dis. 87:775-779.) is a well known and widely applied procedure to prepare clinical samples before inoculation to any type of conventional and rapid mycobacterial culture media including Middlebrook and TK Medium. Phenol red is a component of the solutions used in this procedure but it is not a component of the medium described by Heifets et al. There are no phenol red or any type of pH indicator in the claims of patent US 6,579,694 belonging to Heifets et al.

Specific comments to each "claim rejection"

Regarding the claim rejection according to 35USC&112, it is stated that:

- "Claim 1 and its dependent are drawn to a culture medium however are rendered vague and indefinite for reciting "are configured" in line 6, because the phrase implies that the step is taken to actually configure the inhibitors in a particular way, which is not recited in the claim. It is unclear if steps are actually taken to achieve the claimed configuration, or if the inhibitors intrinsically inhibit growth of non-mycobacterium microorganisms."

In our patent application, in claim 1, it is clearly stated that inhibitors like malachite green that are used to inhibit non-mycobacterial organisms are also used as color dye indicators. Claim 1 states: "A culture medium, comprising: a growth solution, wherein the growth solution is composed of chemicals that promote growth of mycobacteria; one or more indicators, wherein the indicators undergo a spectroscopic change when the pH of the growth solution is altered; and one or more inhibitors, wherein **the inhibitors are configured to inhibit growth of microorganisms that are not mycobacterium, and wherein the inhibitors undergo a spectroscopic change in the presence of mycobacteria.**"

The principle, malachite green working both as an inhibitor and a color dye indicator, is described in detail in the descriptions of our patent application:

"The use of an inhibitor like malachite green that is metabolized by mycobacteria (specifically the strain of mycobacteria of interest) may allow an additional test for the presence of mycobacteria. In such a situation, the inhibitor will gradually be destroyed if the mycobacteria is present. If the mycobacteria is not present, but instead some other type of bacteria was present that induced a spectroscopic change of the indicator, the inhibitor will not be destroyed and thus will indicate that the mycobacteria are not present. In some embodiments a dye (e.g., malachite green) may be used as an inhibitor (i.e., the inhibitor may be a colored compound or exhibit a fluorescence). The use of a dye as an inhibitor may be useful for rapidly assessing the presence or absence of the inhibitor using spectroscopic or visual observation.

The dye malachite green serves as an inhibitor that inhibits the growth of several species of microorganisms other than mycobacteria and makes it selective for mycobacteria. When mycobacteria grow, the dye is metabolized by mycobacteria and the color it gives to the medium is eliminated. The final color by mycobacterial growth is obtained by both the color change of the pH indicator and metabolization of the selective dye. However most species that are resistant to the selective dye can grow in TK Medium and can not metabolize it to eliminate the color it gives to the medium. Thus the final color obtained by the growth of species other than mycobacteria is different than mycobacterial growth. In this way mycobacterial growth can be differentiated from the growth of other contaminant species before confirmation by microscopy or other identification methods. In the present example, the color of the dye is green. When metabolized by the mycobacteria the dye becomes colorless. Therefore, a change in color from red to green indicates that the pH of the medium has changed (thus bacterial growth has occurred), however the inhibitor has not been metabolized. Thus the presence of a green color indicates that microorganisms other than mycobacteria have grown in the medium."

- "Claims 1-4, 6-10 and 12-14 are rejected under 35 U.S.C. 102(b) as being anticipated by Heifets et al. (US 2001/0055787 A1).

Ms. Davis states, "Heifets teaches an agar culture medium for mycobacterium tuberculosis, wherein the medium comprises 7H10 agar, polymixin B, amphotericin B, trimethoprim, and antimicrobial drugs. The medium comprises growth serum, glycerol, salts and phenol red."

As described above in our general comments, the statement above is not true. The medium described by Heifets et al. does not include phenol red or any other color dye pH indicator. Phenol red is only mentioned in the descriptions as a component of a solution used in the decontamination and concentration procedure described by Kubica et al. (Kubica GP, Dye WE, Cohn ML, Middlebrook G. Sputum digestion and decontamination with N-acetyl-L-cysteine-sodium hydroxide for culture of mycobacteria. 1963. Am. Rev. Respir. Dis. 87:775-779.) which is used to process the clinical samples before inoculating to any type of mycobacterial culture medium. The most important modification of Middlebrook 7H10 medium described by Heifets is the addition of growth serum which is not a component of TK Medium described in our patent application. Therefore the reference does not anticipate the claimed subject matter and the claims 1-4, 6-10 and 12-14 of our patent application can not be rejected based on the patent application of Heifets et al.

- "Claims 1-3 and 7-8 are rejected under 35 U.S.C 102(b) as being anticipated by Miyamoto et. al. (US 6197574 B1)."

Ms Davis states, "Miyamoto teaches a culture medium comprising l-lysine, glucose, ammonium iron, magnesium chloride, bromocresol purple, and malachite green."

In his/her application, Miyomoto claims an apparatus that can follow the growth of microorganisms and states that the medium that will be used in this apparatus will be selected from one of four different media which are designed to grow, Salmonella, Vibrio parahaemolyticus, Escherichia coli group and Staphylococcus aureus, which cause food poisoning. None of these media have the ability to grow mycobacteria since they lack essential elements like albumin which are vital for mycobacteria. The components of the said media are different than TK Medium. TK medium contains egg as the albumin source. It has both the ability to grow mycobacteria and indicate growth by color change. Additionally TK Medium has the ability to differentiate the growth of mycobacteria from the growth of other contaminating organisms. Since the composition and function of TK medium is completely different than the media claimed by Miyomoto et al. Claims 1-3 and 7-8 of our application can not be rejected based on the claims of Miyomoto et al.

- "Claims 14 - 5 and 11 are rejected under 35 U.S.C. 103(a) as being unpatentable over Heifets."

Ms Davis states, "Heifets does not teach the medium wherein the gelling substance is an egg solution or wherein each of the claimed antimicrobial compounds are included. However, at the time of the claimed invention, egg based gelling substances were standard alternatives to agar in the art and were routinely used in culture media. Thus it would have been obvious to one of ordinary skill in the art to use an egg based medium in the culture of Heifets as a matter of standard practice."

As described above, the medium described by Heifets et al. is different in composition and in working principle than TK Medium. So TK Medium can not be considered simply as "egg based medium in the culture of Heifets." The reference does not anticipate the claimed subject matter and the Claims 14 - 5 and 11 can not be rejected based on the claims of Heifets et al.